Radioactivity - Radionuclides - Radiation
8th Nuclear Science Training Course with Nuclides.net
(Institute Jožef Stefan, Ljubljana, 13th-15th Sept. 2006)

Thursday, 14th Sept. 2006

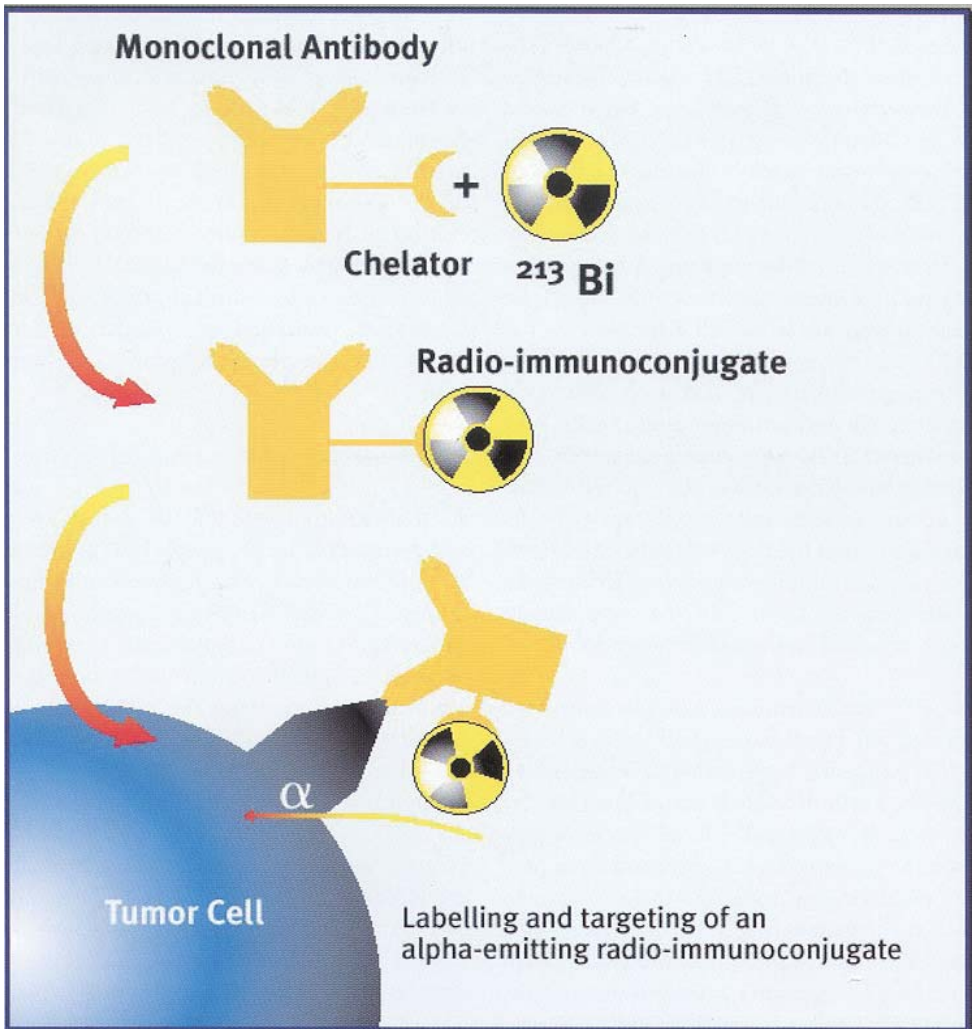
Radio-Immunotherapy with Alpha-Emitters

Alfred Morgenstern
European Commission
Institute for Transuranium Elements
P.O. Box 2340, 76125 Karlsruhe, Germany
E-mail: Alfred.morgenstern@ec.europa.eu

Overview:

- Principle of Alpha-Immunotherapy
- Production of Ac-225 / Bi-213
- Pre-clinical and clinical studies

I. Principle of Alpha-Immunotherapy



Specific recognition of cancer cells

Vector/Carrier

(e.g monoclonal Ab's, Ab-fragments, peptides)

+

Chelate/Linker

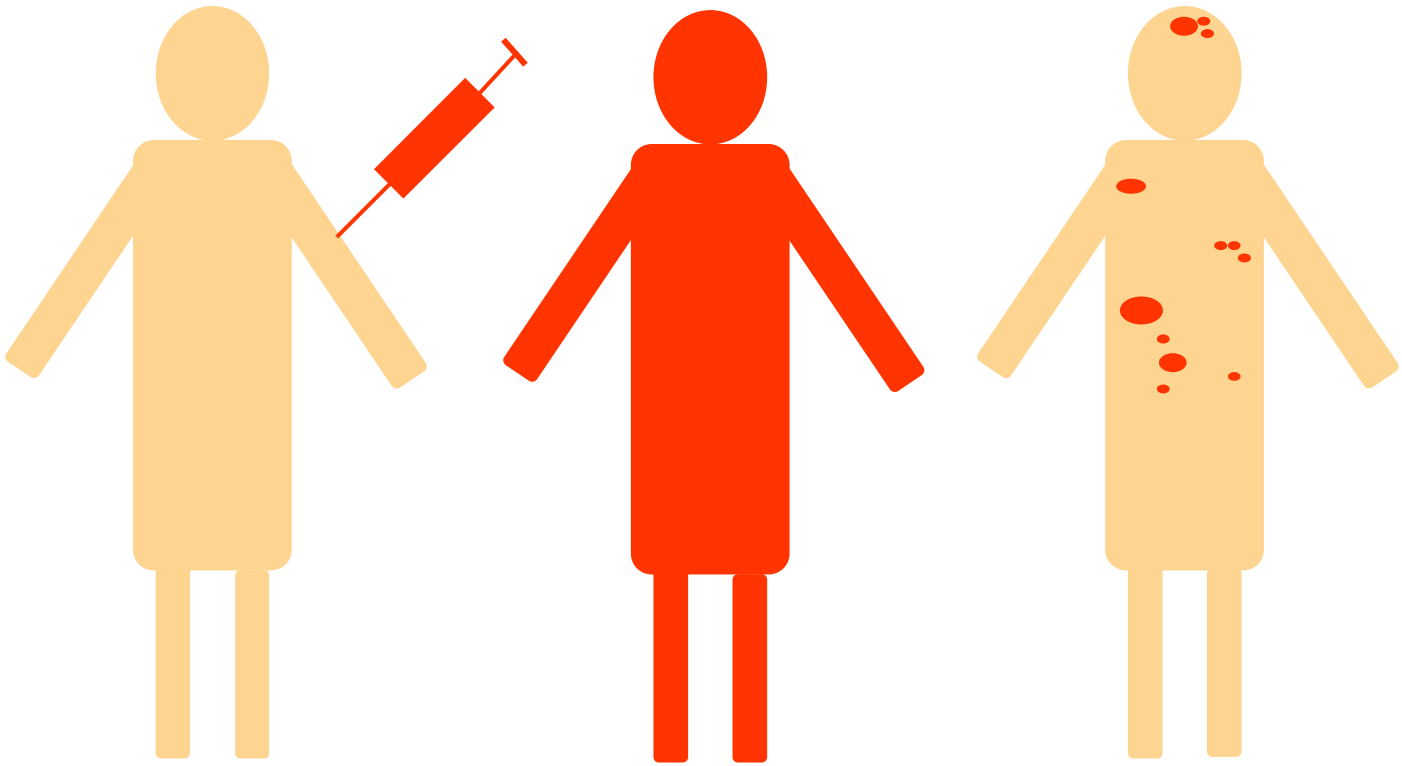
(e.g. derivatives of DTPA, DOTA)

+

Effective killing

α - Emitters

(e.g. Ac-225, Bi-213, At-211)

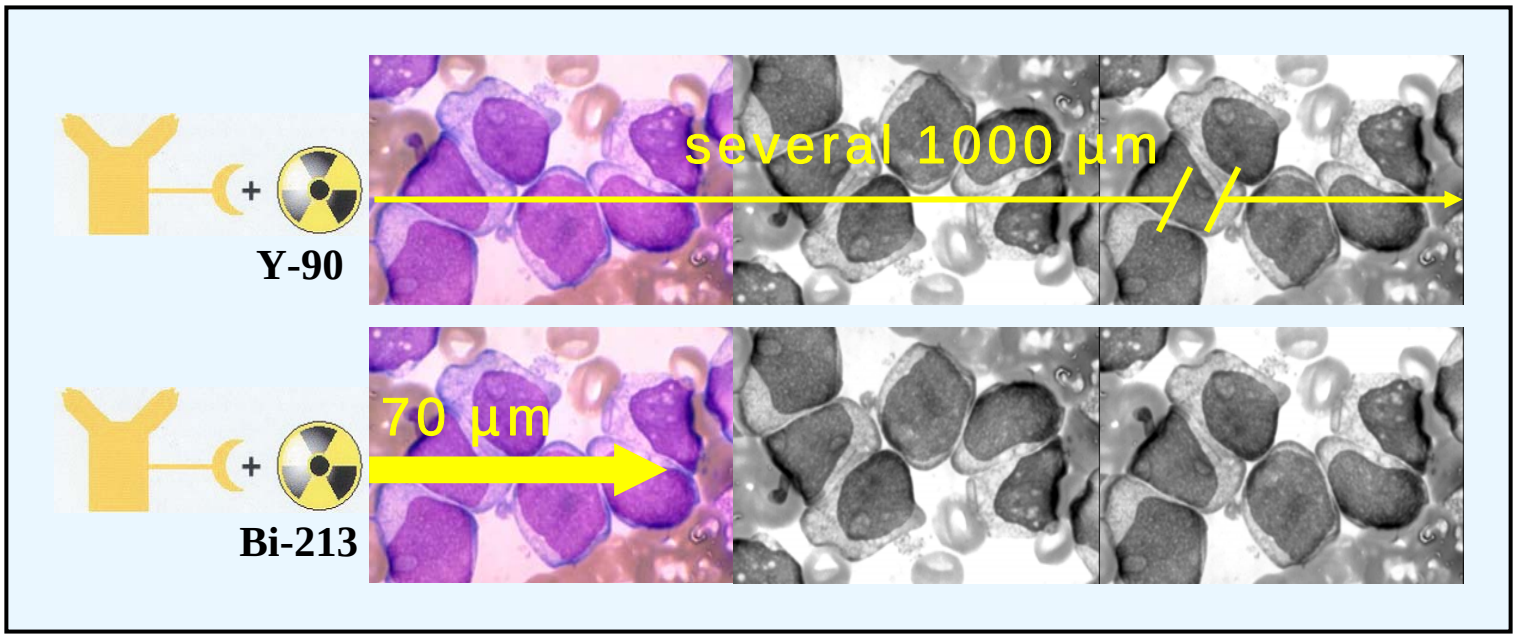


Injection **Circulation** **Localization**

Whole body distribution

Radioimmunotherapy β and α

Beta particles:
Energy: 0.1 - 1.5 MeV, Range in soft tissue: up to several mm's
Corresponding number of cells: 10-1000



Alpha particles:
Energy: 4-9 MeV, Range in soft tissue: 40-90 µm
Corresponding number of cells: 2-10

Linear Energy Transfer (α) ≈ 100 x LET (β)

Joint Research Centre

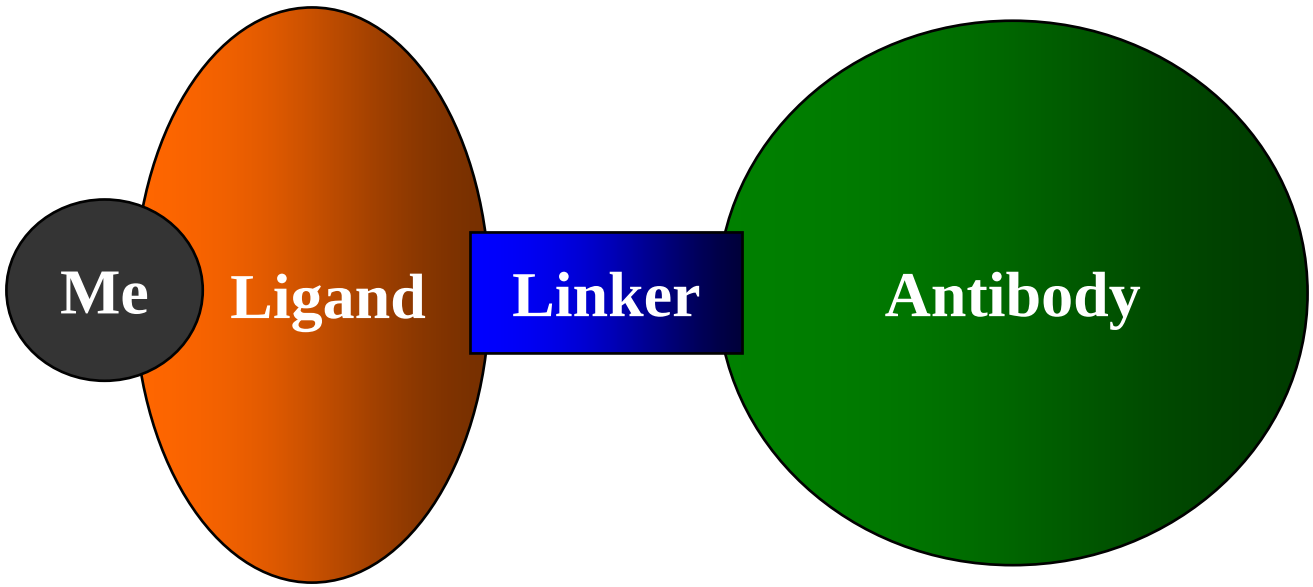
Possible applications of Alpha-Immunotherapy:

- Monocellular cancers (e.g. Leukemia)
- Micrometastatic disease
- Minimal residual disease after chemotherapy
- Residual tumor tissue after tumor resection (e.g. brain tumors)
- Bacterial, viral and fungal infections in patients with impaired immune systems

Multidisciplinary approach

Anorganic Chemistry
and Radiochemistry

Molecular Biology



Organic Chemistry

Joint Research Centre

ITU's main activities in the field of Alpha-Immunotherapy

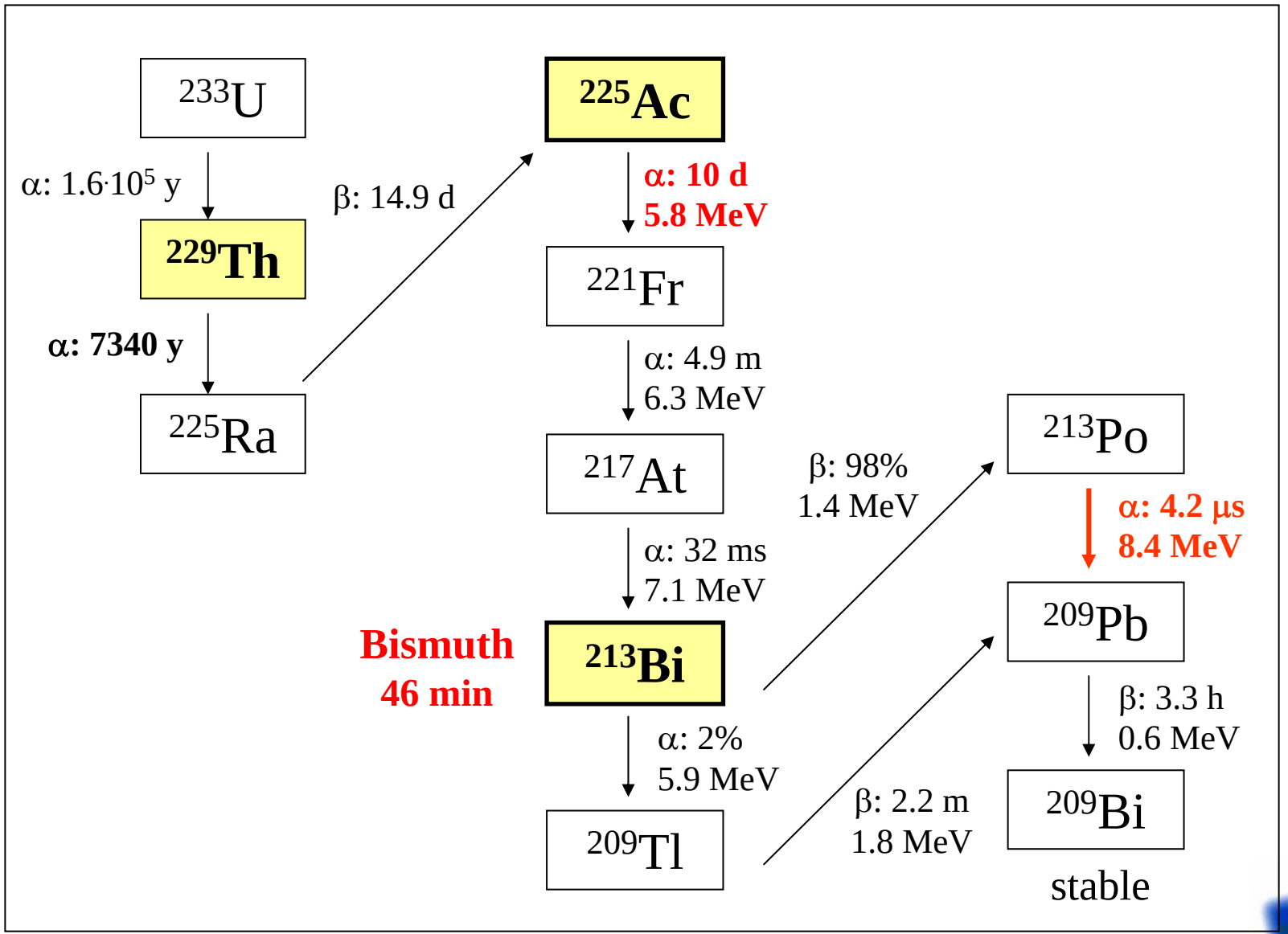
- Production of alpha-emitters and radionuclide generators
- *in vitro* testing
- Support pre-clinical research and clinical trials
- Ensure safe use in hospitals:
  provision of radionuclide generators, equipment, training
- Labelling of biomolecules: chelation, chelate development
- Radiobiology: mechanisms of radiation induced effects in cells

Potential alpha emitters for AIT:

Isotope	Half-life	Production routes	Limitations
Tb-149	4.1 h	Pr-141(^{12}C,4n)Tb-149 Nd-142(^{12}C,5n)Dy-149 => Tb-149 spallation (Ta)	accelerator produced, short half-life
Bi-212	1.0 h	Th-228	⇨ Tl-208 high energetic γ-emitter
Ra-223	11.4 d	Ra-226(n,γ)Ac-227	chelate
Ra-224	3.66 d	Th-228	chelate
At-211	7.2 h	Bi-209(α,2n)At-211	cyclotron produced, labelling, stability *in vivo*
Ac-225	10.0 d	I. U-233 =>Th-229 II. Ra-226(p,2n)Ac-225	chelate stability *in vivo,* toxicity of daughter nuclides
Bi-213	45.6 min	Ac-225	limited availability from Th-229

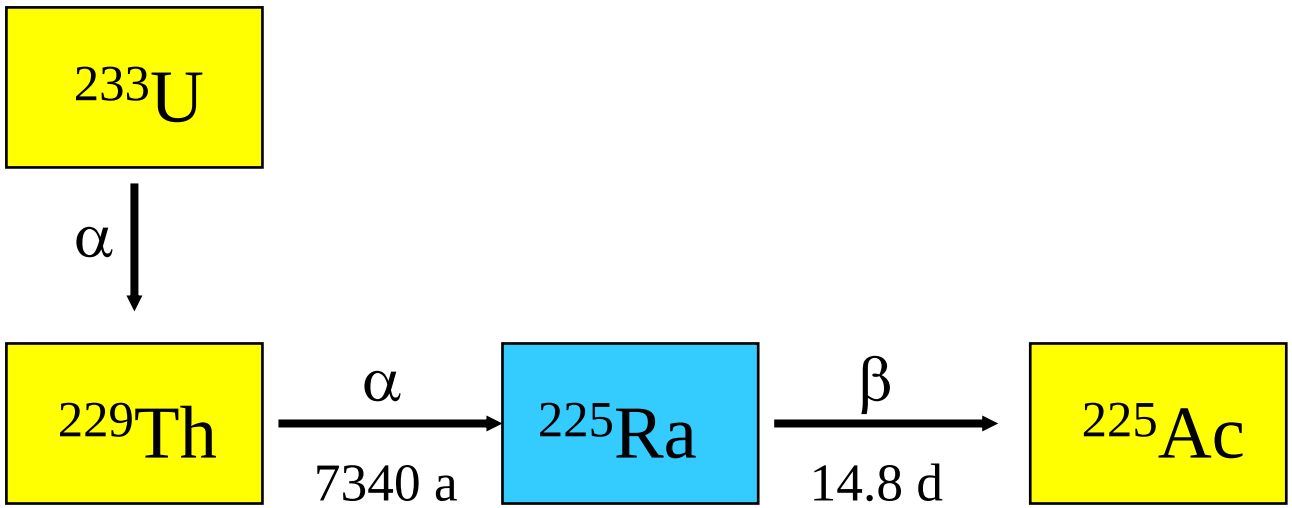
Joint Research Centre

The Ac-225/Bi-213 system



Joint Research Centre

Ac-225/Bi-213 Production:
I. Separation of Ac-225 from Th-229

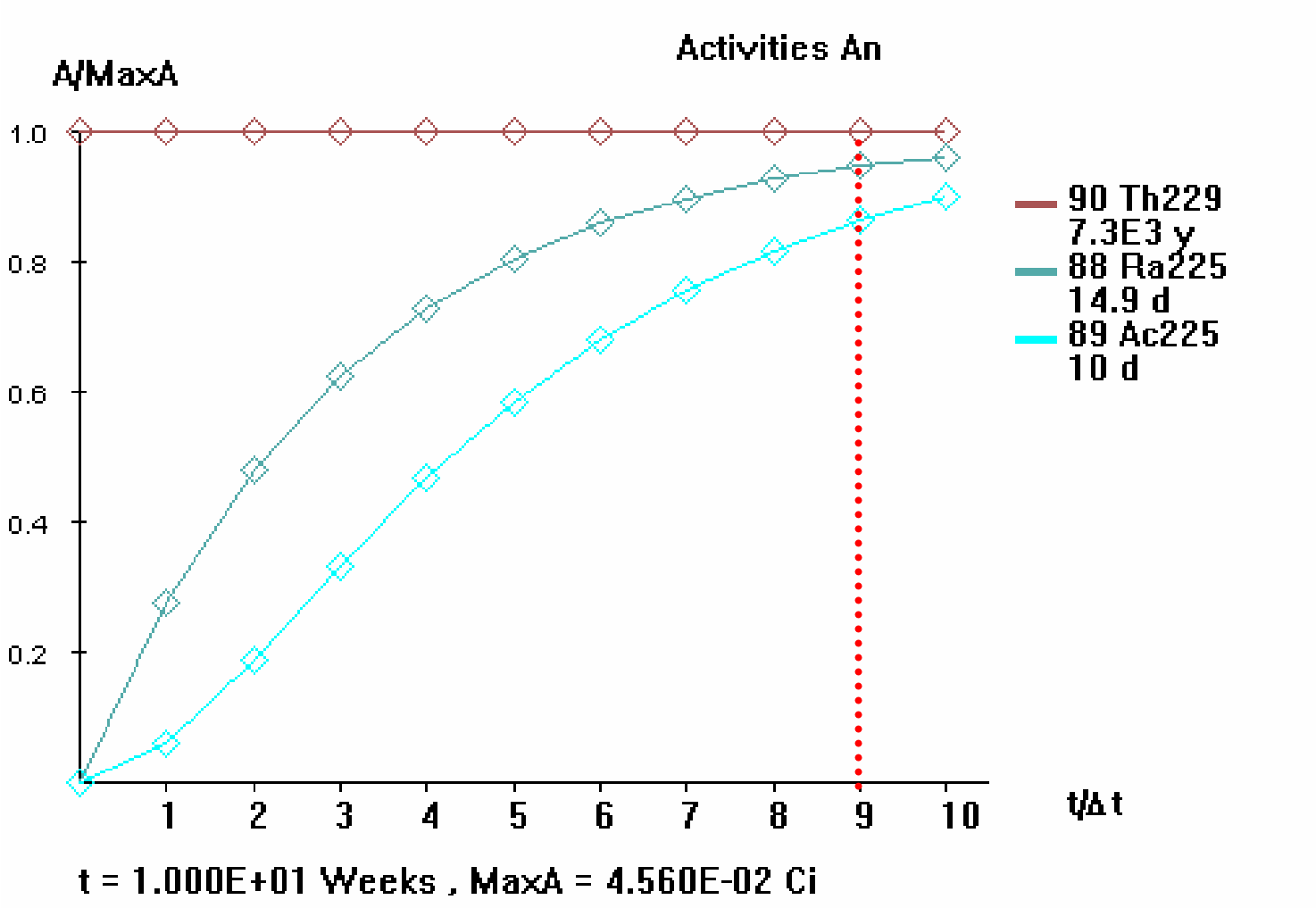


- ITU's Th-229 source: 45.6 mCi
  => every 9 weeks 43.2 mCi Ra-225 and 39.4 mCi Ac-225 available
  => provided to partners free-of-charge on the basis of scientific collaborations

- Only 2 other Th-229 sources exists worldwide:
  Oak Ridge National Lab, approx. 150 mCi,
  IPPE Obninsk, approx. 20-30 mCi
  current market price: approx. 1000,- to 1500,- Euro / mCi Ac-225

Joint Research Centre

Build-up of Ra-225, Ac-225 from Th-229



Joint Research Centre

Analytical challenges

- Th-229 source: dose rate approx. 100 mSv/h
- Separation of Ac-225:

  rapid (Ac-225: $T_{1/2}$ = 10 d)

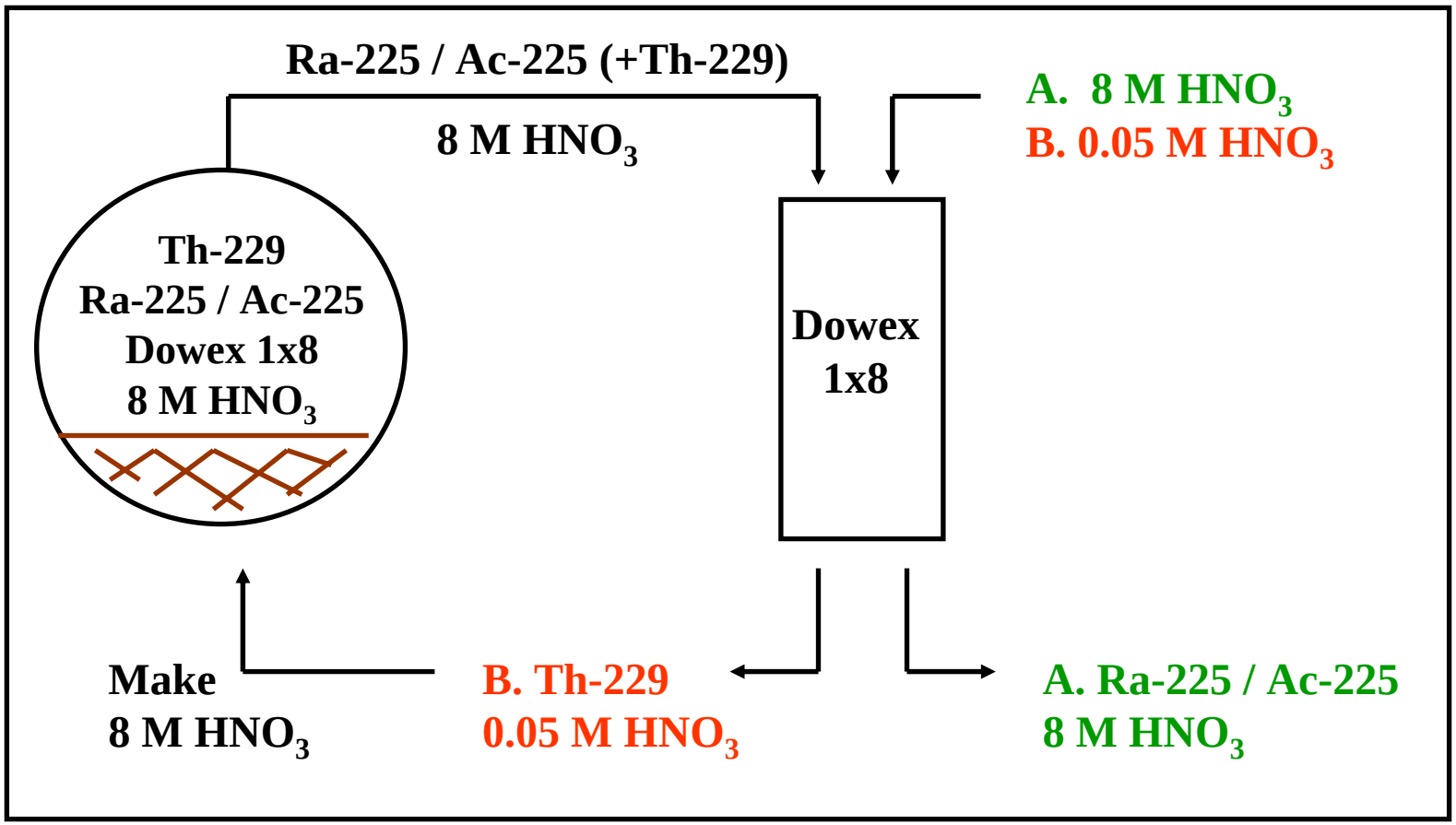
  simple and robust process suitable for glove box environment

  high yield of Ac-225

  high purity, clinical grade product
- no loss of Th-229

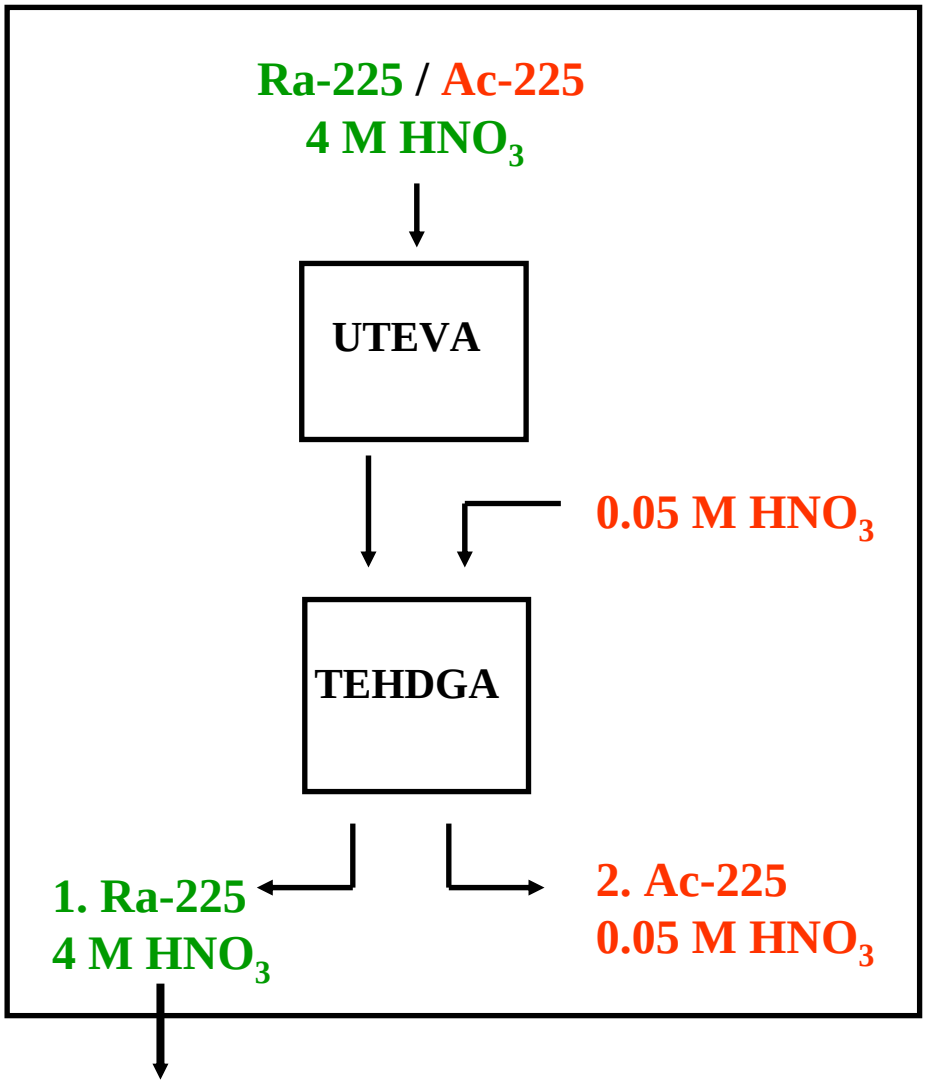
Joint Research Centre

Th-229 / Ra-225, Ac-225 separation: Anion exchange



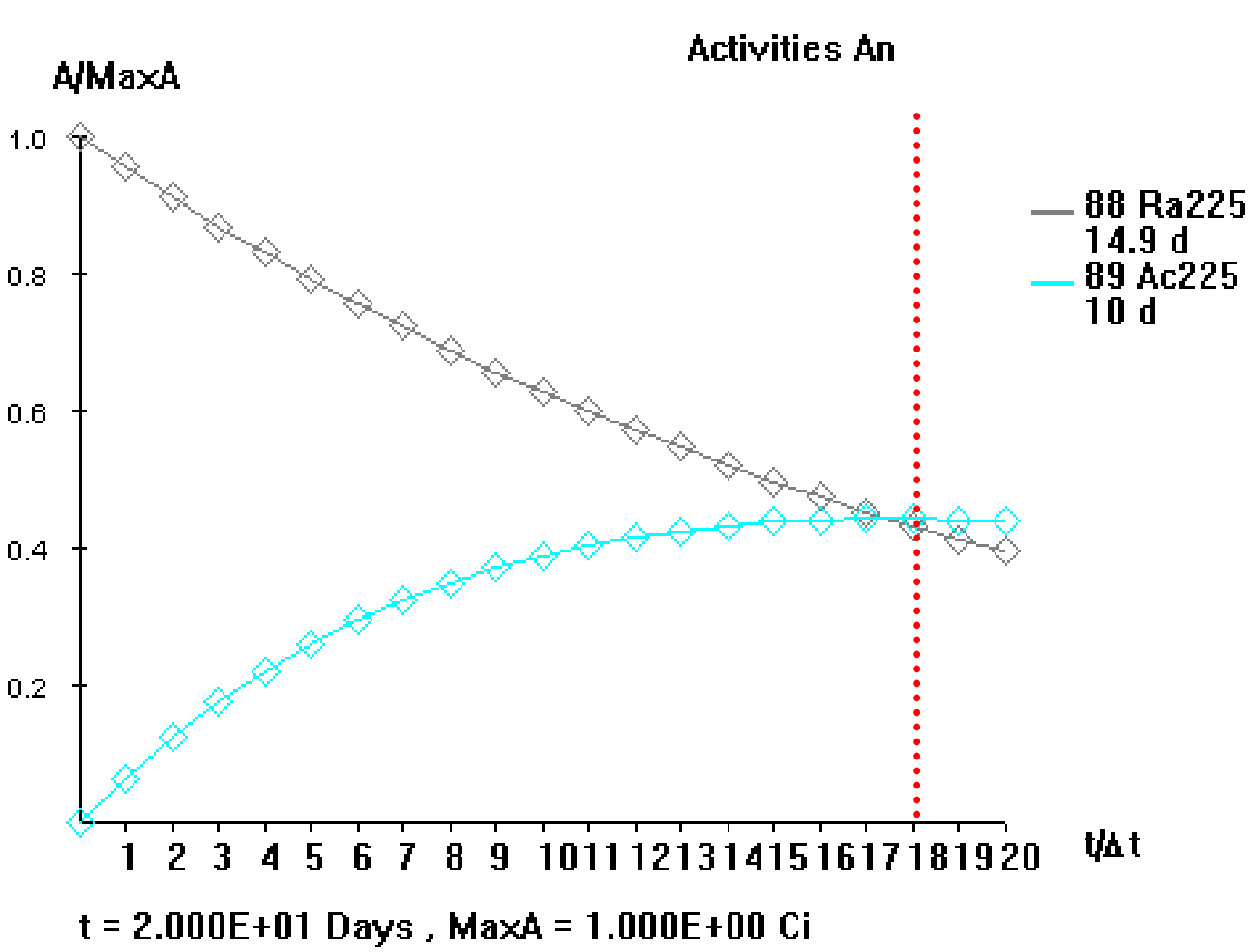
Joint Research Centre

Ra-225 / Ac-225 separation: Extraction chromatography



Joint Research Centre

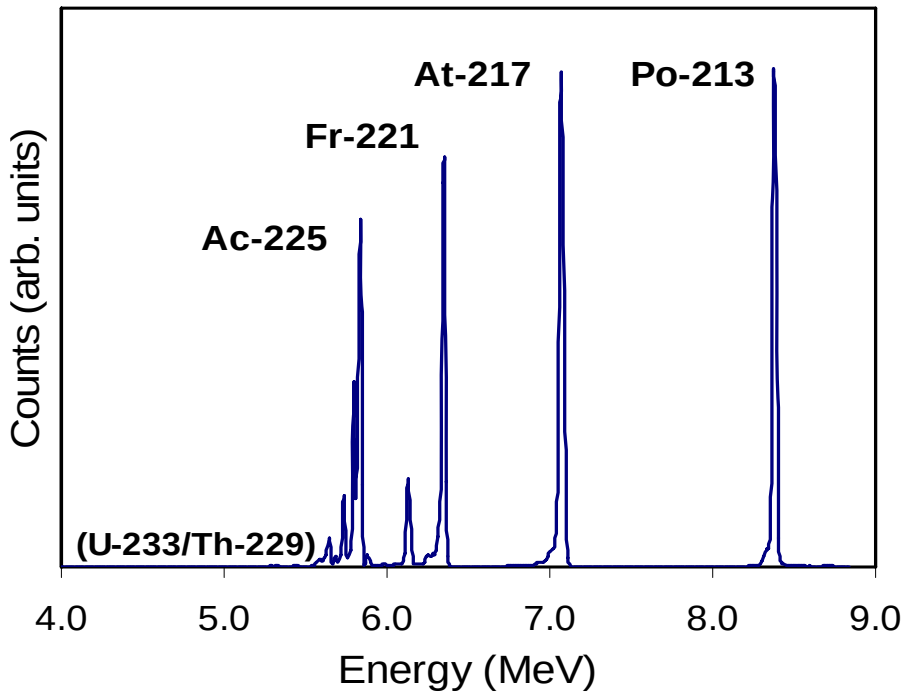
Ac-225 build-up from Ra-225 decay

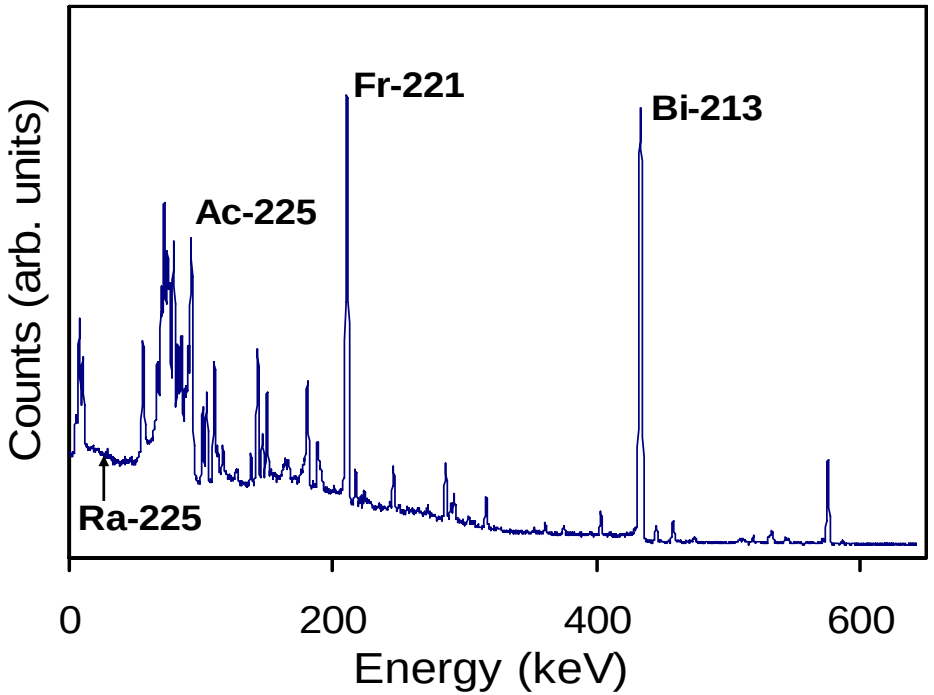


Joint Research Centre

Quality control:

- Alpha-, gamma-spectrometry
- ICP-MS

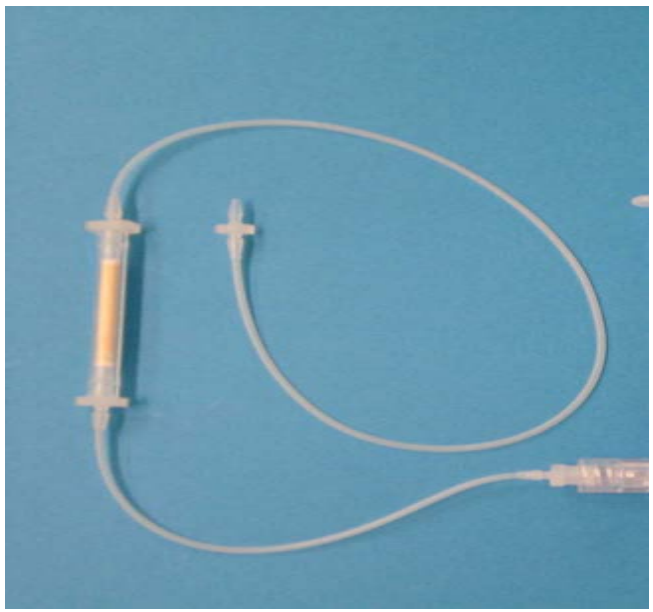


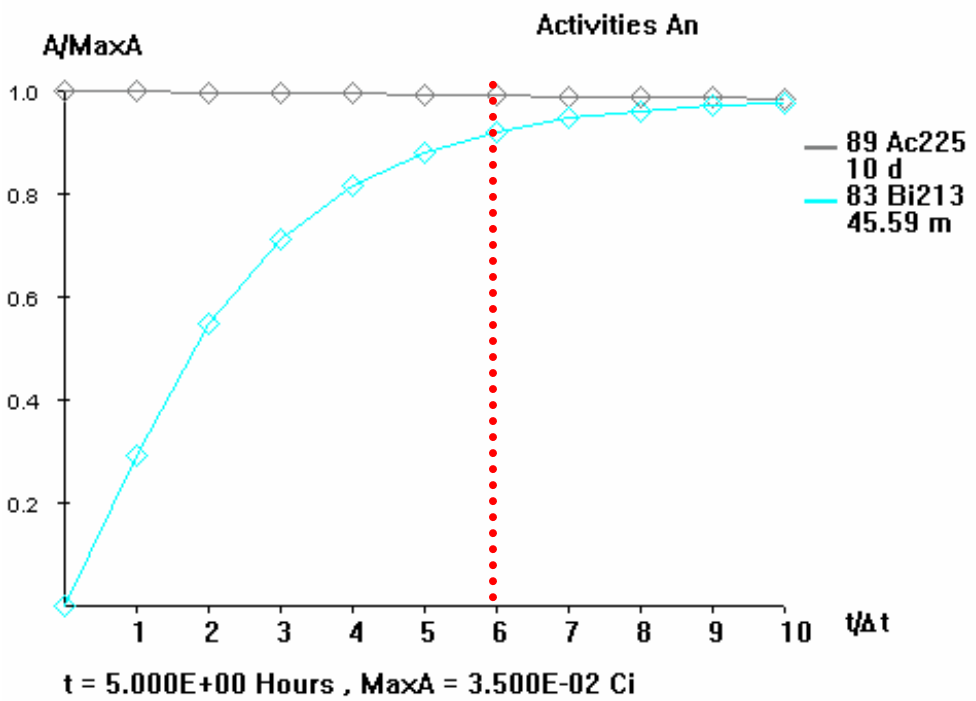


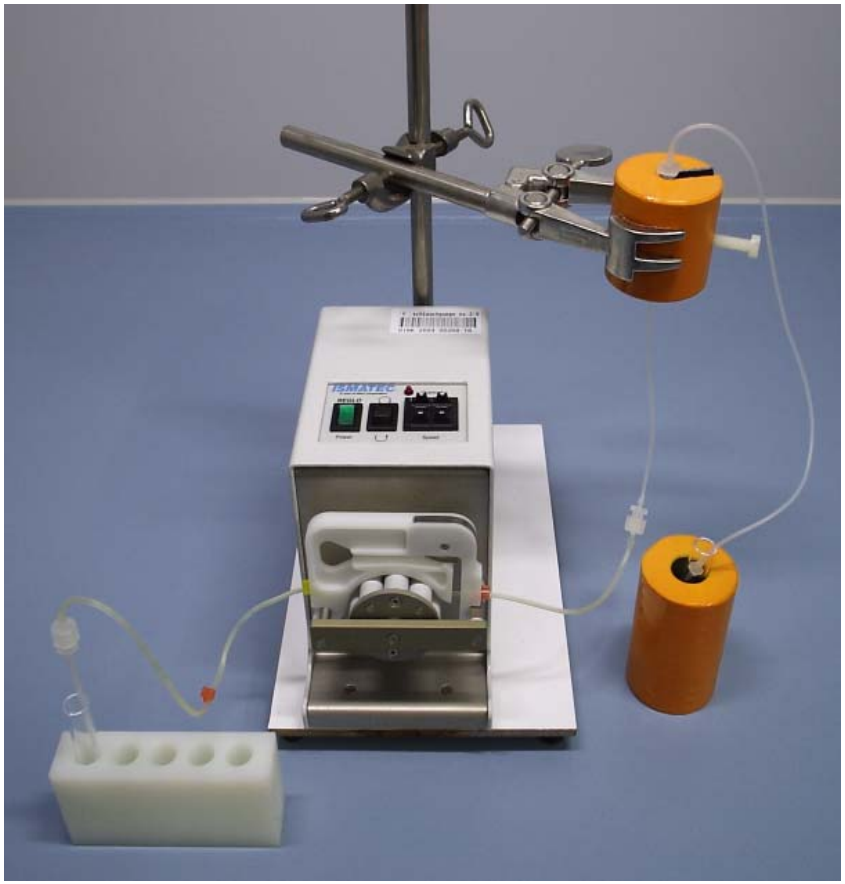
Overall yield of separation process: > 98%

Joint Research Centre

Generator loading and shipment







Shipments from ITU until today:
> 200 generators
=
approx. 3.8 Ci Ac-225

II. Cyclotron production: Ra-226(p,2n)Ac-225

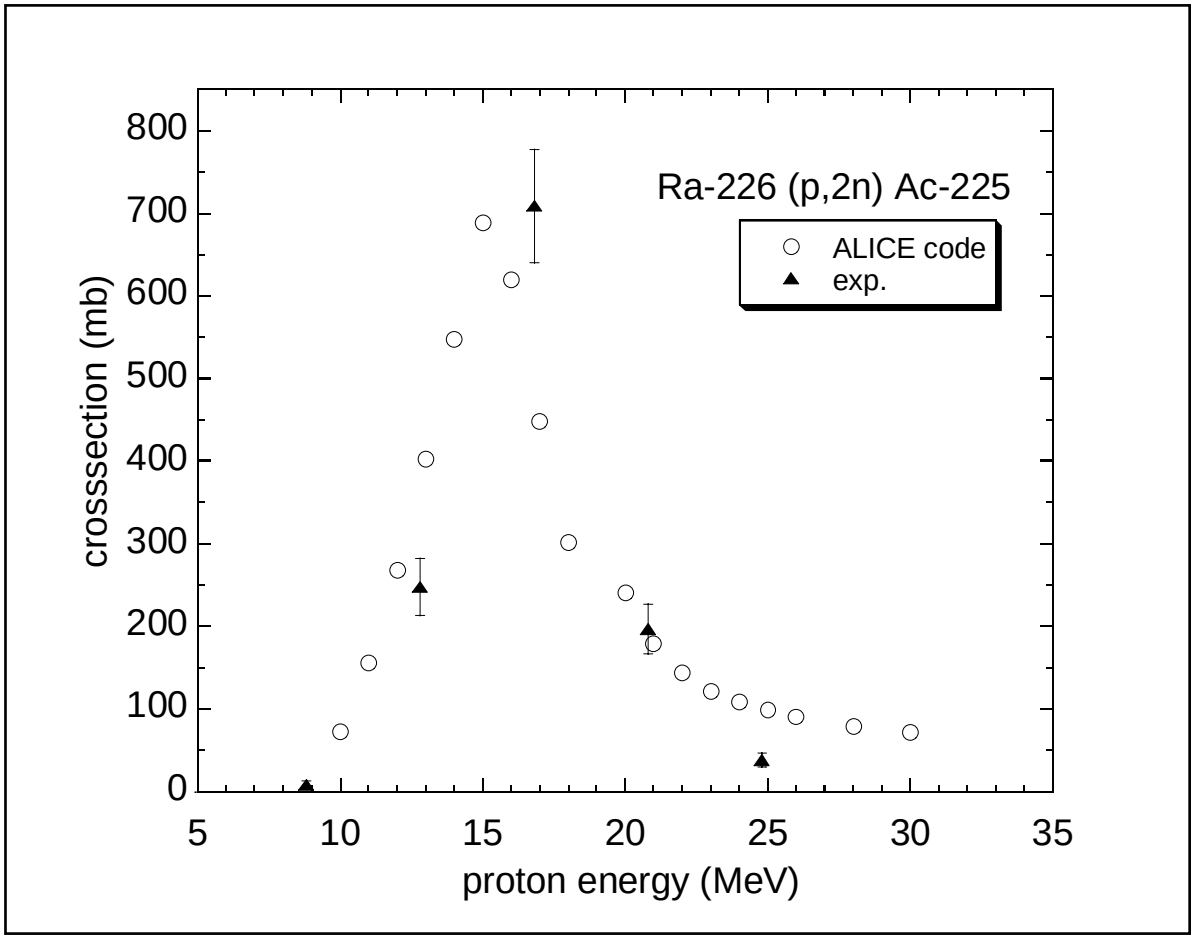
1. Determination of activation cross-sections as function of proton energy

2. Demonstration of feasibility of large-scale production

Determination of activation cross-sections:

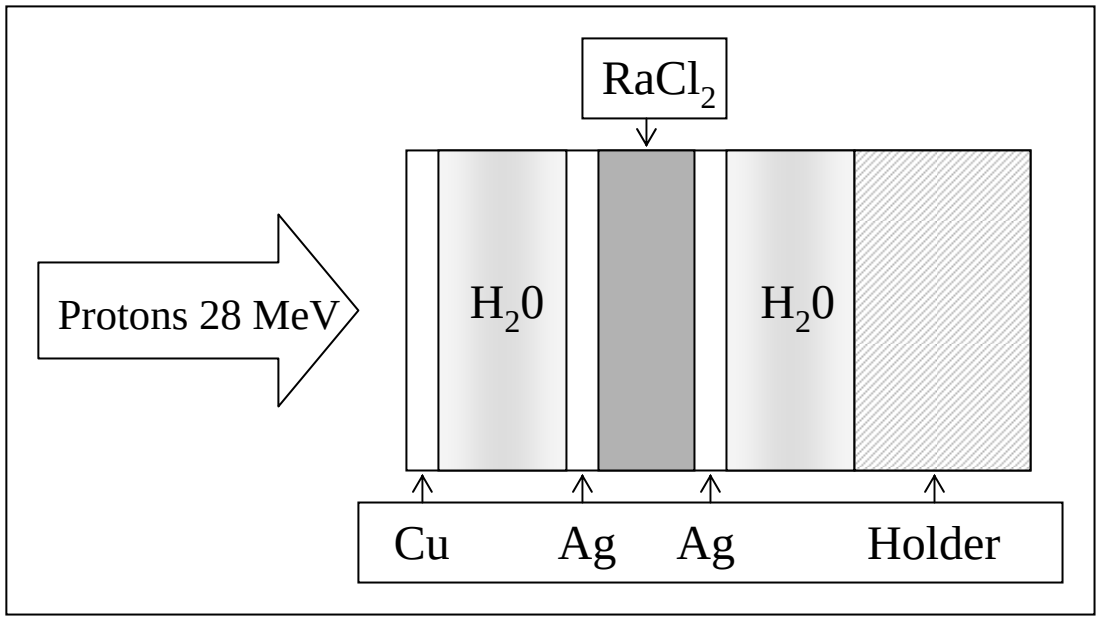
Irradiation parameters:

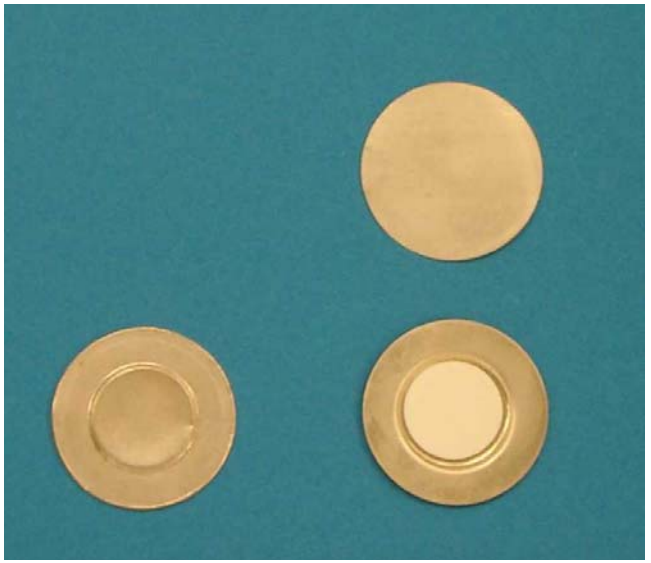
- 12.5 µg Ra-226 per sample
- thin targets
- 28 MeV protons, 7 h, 10 µA
- silver foils of varying thickness for energy attenuation
- incident proton energies 8.8 - 24.8 MeV



=> experimentally determined cross-sections agree well with model calculations (ALICE code, LLNL)

Demonstration of large-scale production



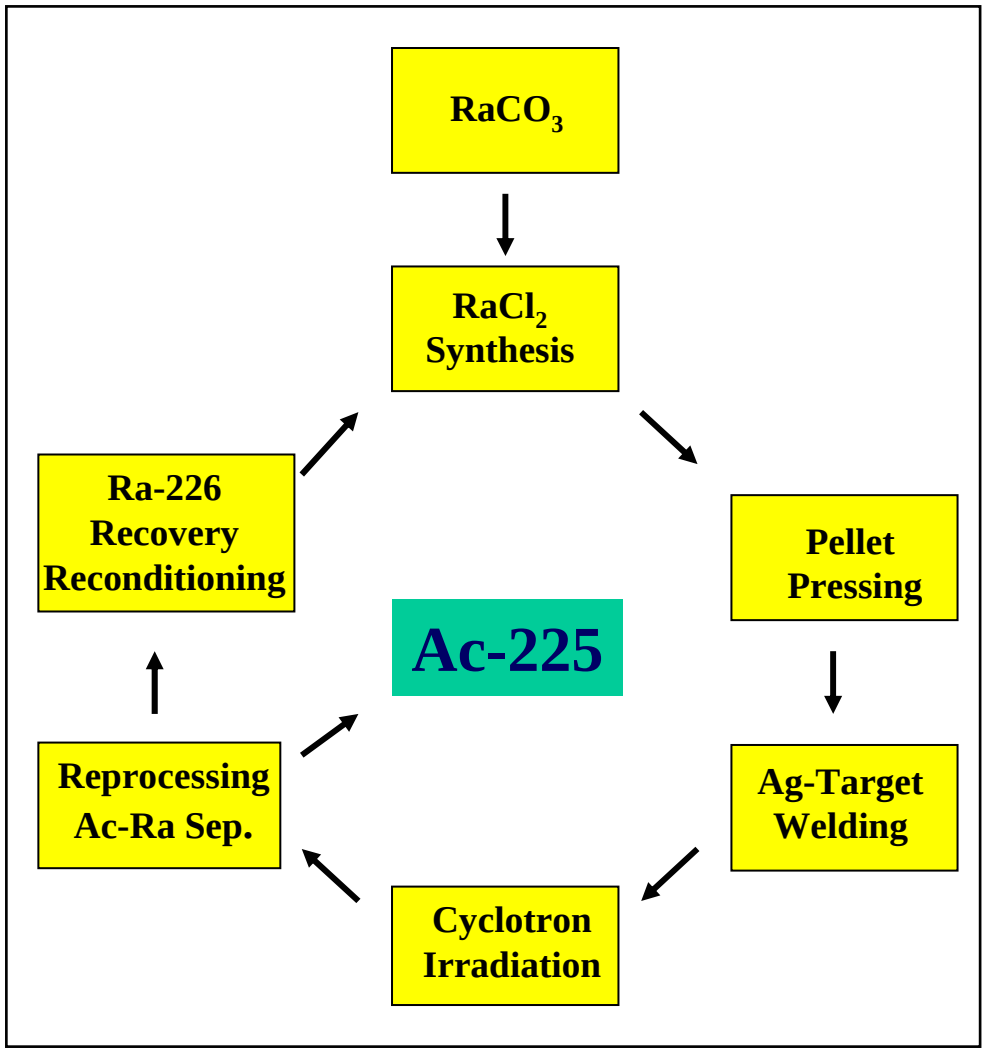


Targets:

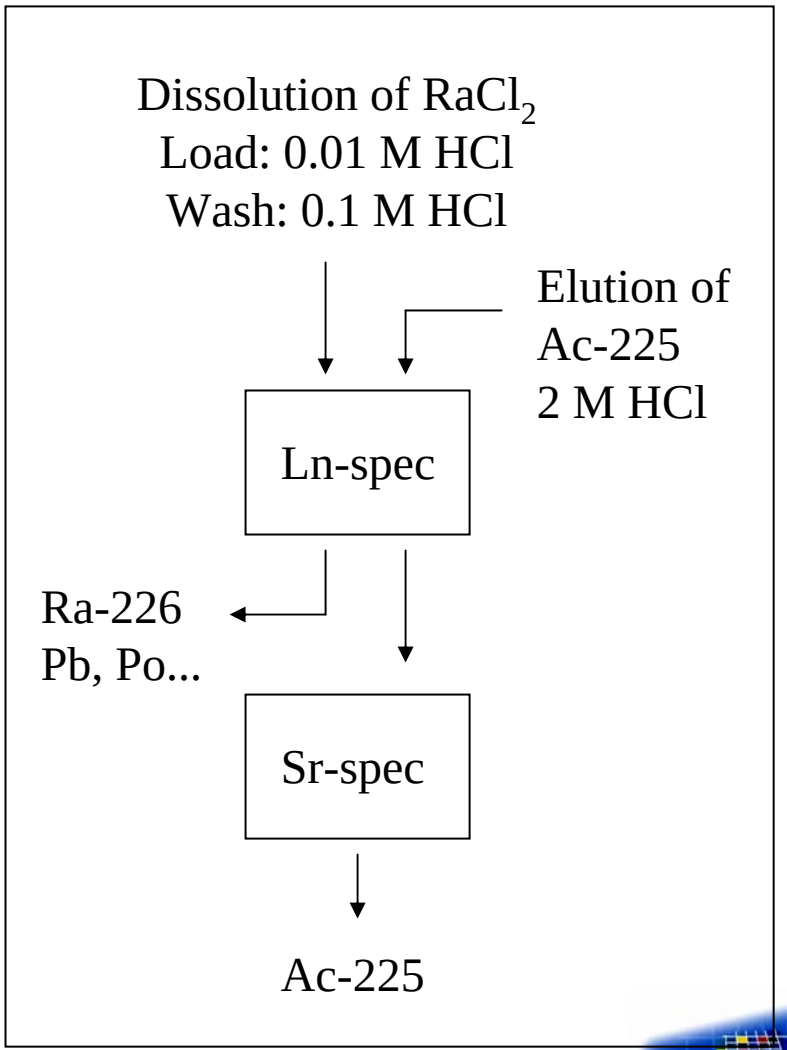
- 30 mg Ra-226 in 300 mg $BaCl_2$ matrix
- extensive target testing before irradiation
- on-line monitoring of alpha activity in cooling circuit

Joint Research Centre

Irradiation cycle



Ac-225 purification



Results of production runs

Mass of Ra-226 (mg)	30.0	30.5	30.1
Proton current (µA)	25 / 50	20	50
Incident proton energy (MeV)	16.0	16.0	15.9
Target surface (mm^2)	200	200	200
Irradiation time (h)	49.3	26.3	45.3
Ac-225 / Ra-226 (% activity)	12.5	18.7	43.5
Ac-225 produced (mCi)	3.8	5.7	13.1

Quality control:
- Radiochemical purity: alpha, gamma spectrometry
- Bi-213 labelling studies

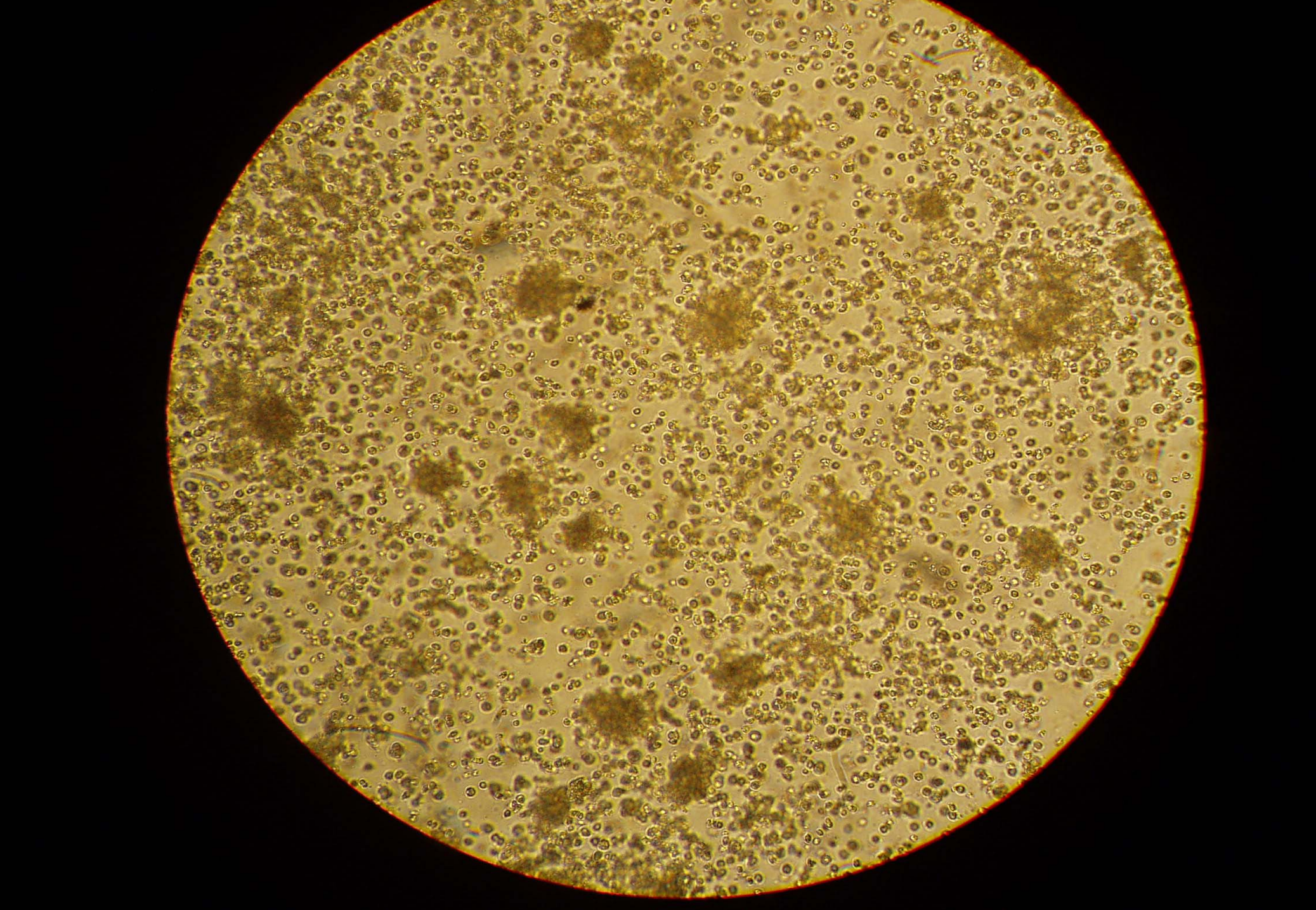
=> no significant differences between Ac-225 from Th-229 route and from Ra-226

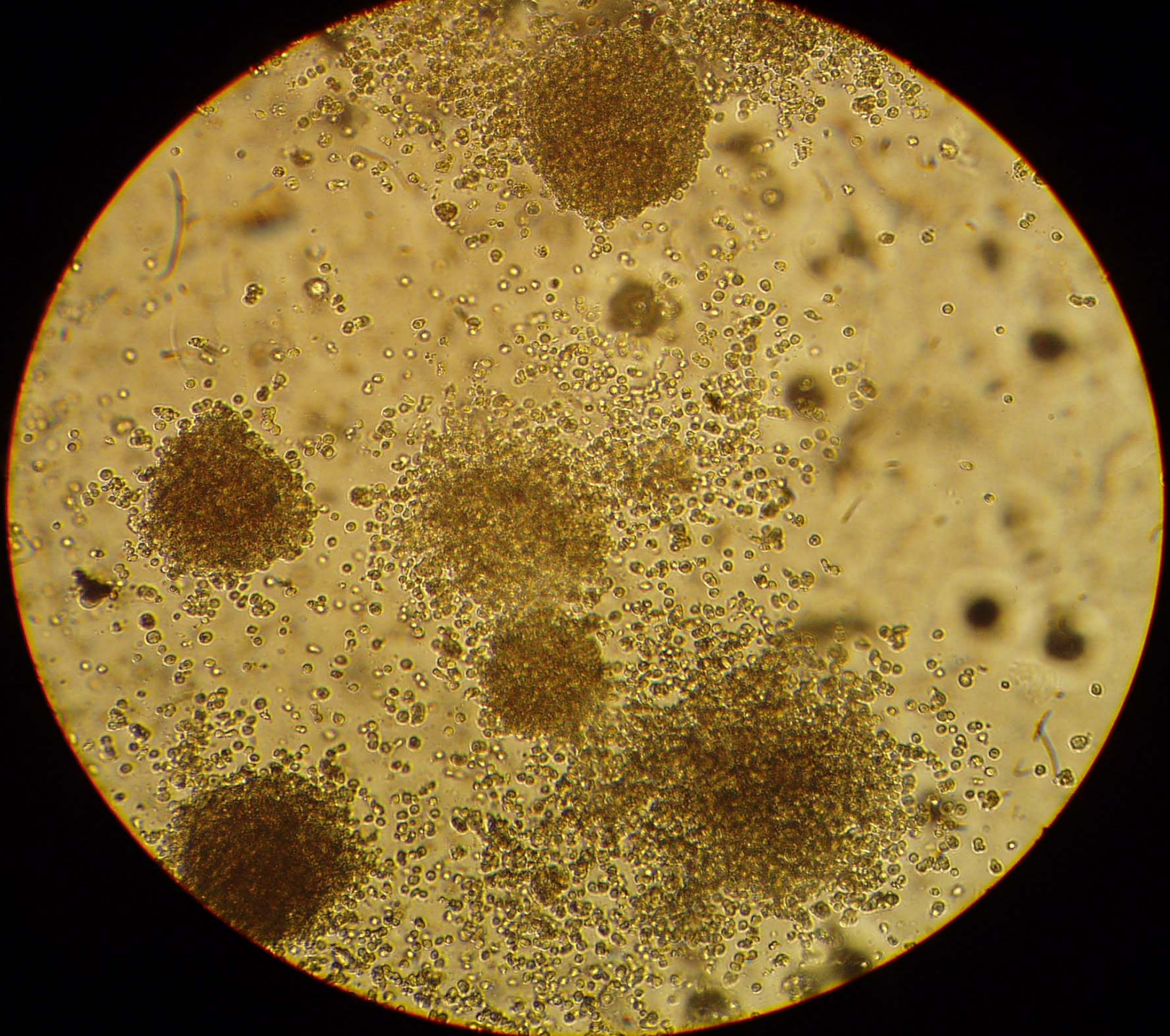
Pre-clinical studies
In vitro testing

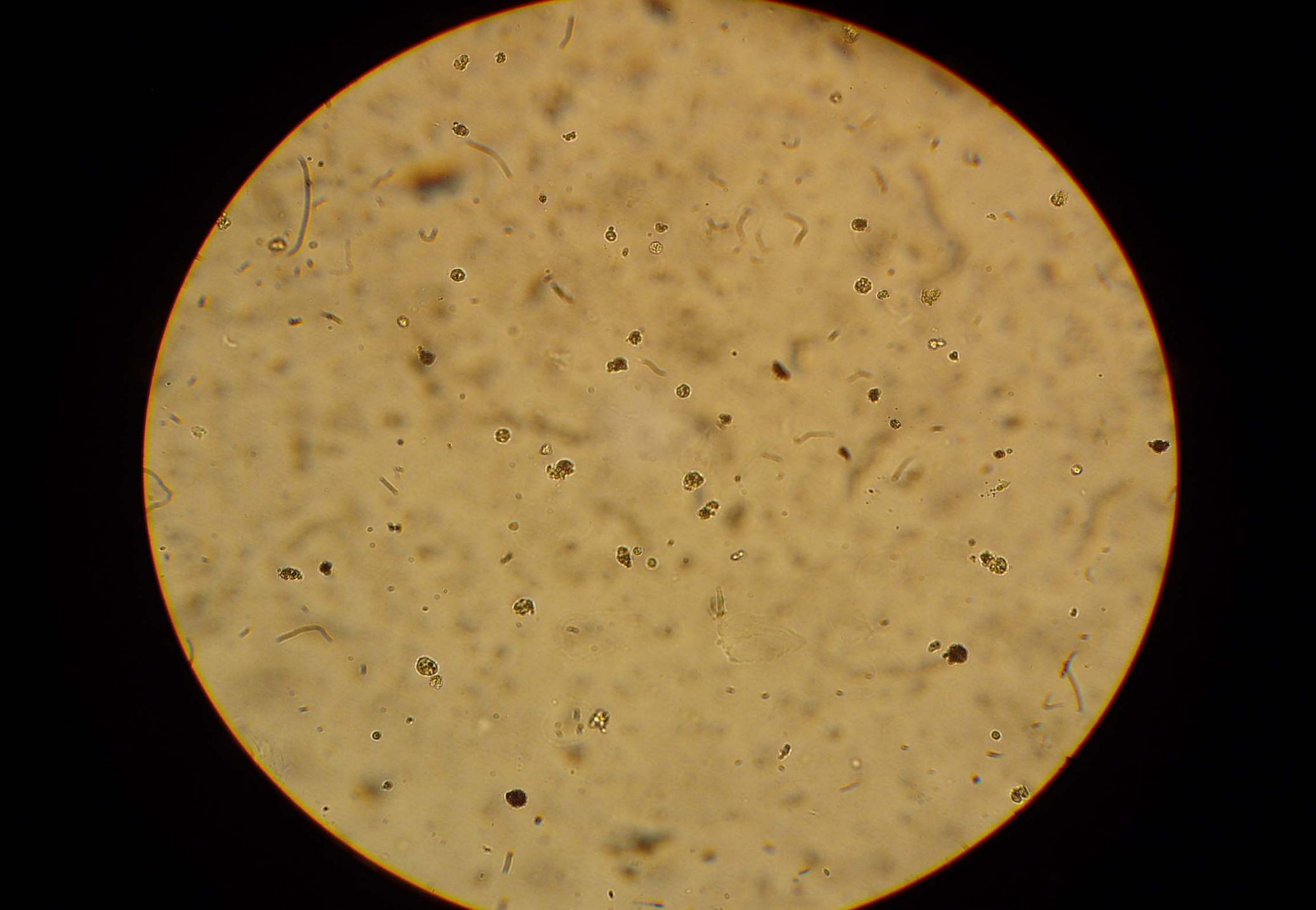
e.g.: Colony forming assay:

Colony forming tumour cells are incubated with Bi-213 labelled specific antibodies

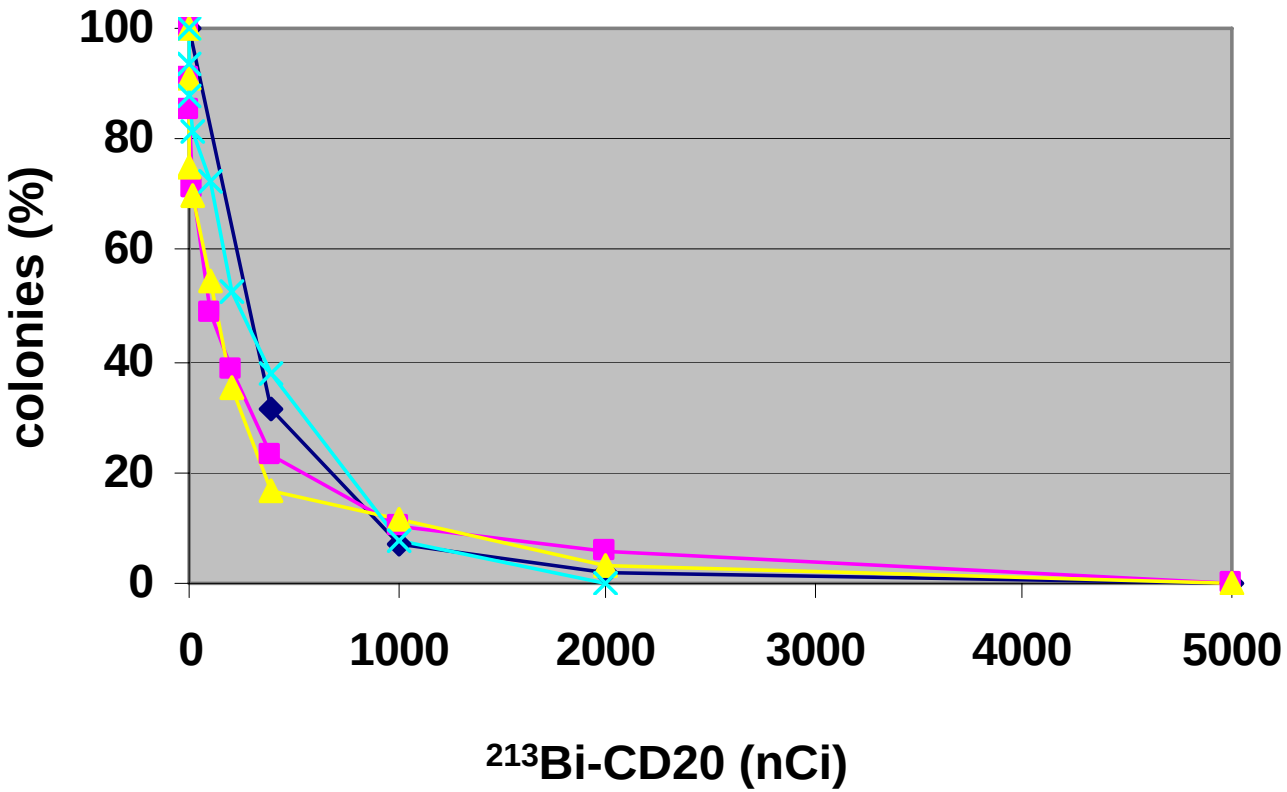
Ability to form tumour colonies is compared with untreated controls



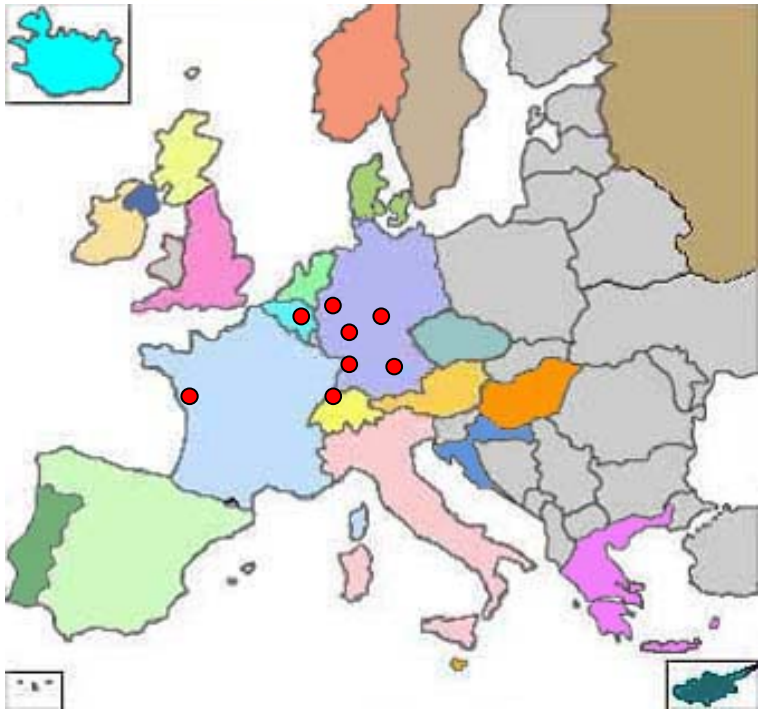




Colony forming assay of K422 lymphoma cells with Bi-213 labelled anti-CD20 antibody



Pre-clinical and clinical collaborations (1)



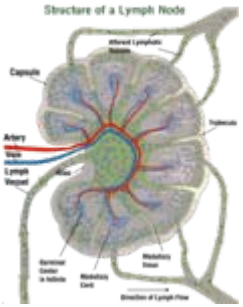
Europe

Germany	**Heidelberg**
	Düsseldorf
	München
	Ulm
France	**Nantes**
Belgium	**Gent**
Switzerland	**Basel**

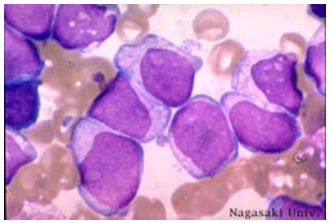
USA, Australia

USA	**MSKCC, New York**
	AECM, New York
	GSCC, New York
	NIH, Maryland
Australia	**Sydney**

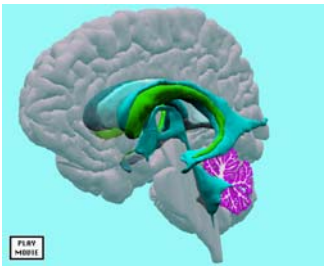
Selected collaborations (2)



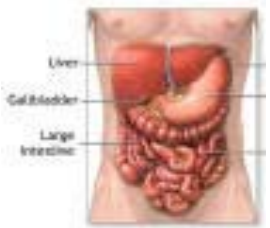
Lymphoma
(Heidelberg, Düsseldorf)



Acute leukaemia
(Ulm, New York)

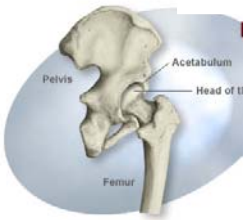


Brain tumors
(Basel)

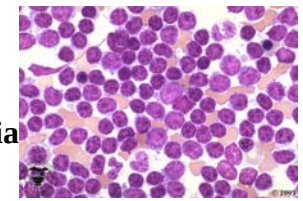


Gastric cancer
(Munich)

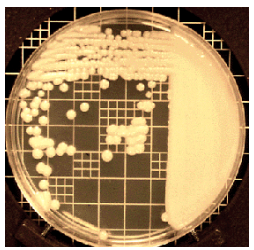
ITU
α-emitting isotopes



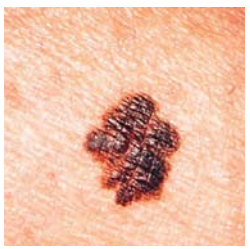
Multiple myeloma
(Nantes)



Chronic leukaemia
(Gent)



Infectious diseases
(New York)



Malignant melanoma
(Sydney)

Joint Research Centre

Pre-clinical studies

ITU - Technical University Munich

Gastric Cancer

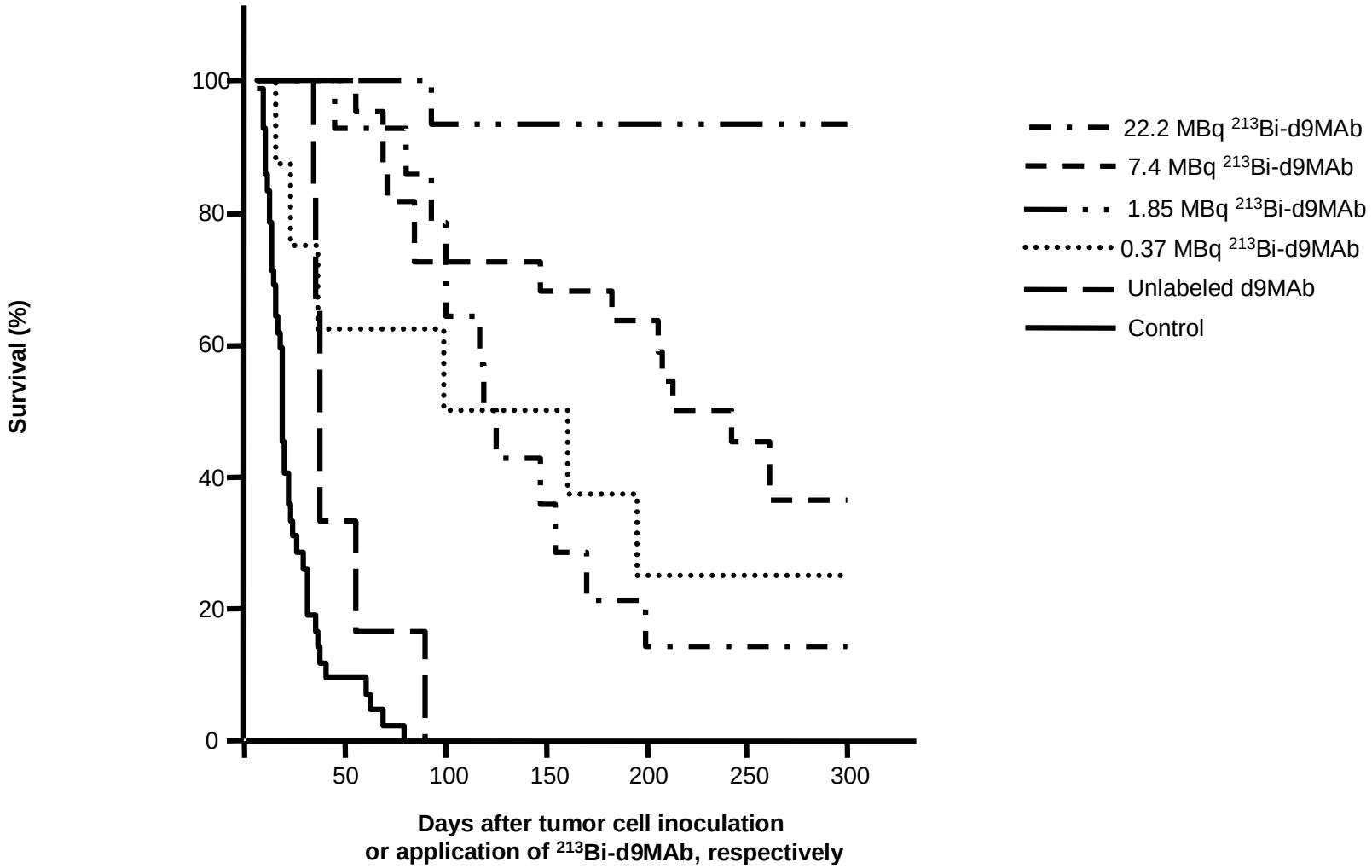
Problem: High tumor recurrence even after tumor resection because of early tumor cell dissemination => metastases

Animal model

- Inoculation of HSC45-M2 cells (tumour cells) i.p. in 6-week old mice
- At 24 h, 8 d and 15 d after tumor cell inoculation, treatment of animals (once / twice) with different activities of ^{213}Bi-d9MAb (0.37 – 22.2 MBq)
- Locoregional application

Joint Research Centre

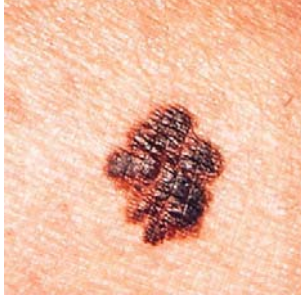
Therapeutic Efficacy : Survival of xenotransplanted mice after locoregional ^{213}Bi-immunotherapy



R. Beck, C. Seidl, B, Pfost, A. Morgenstern, F. Bruchertseifer, M. Schwaiger, R. Senekowitsch-Schmidtke, Clinical Cancer Research 2006, submitted

Joint Research Centre

Clinical trials



Phase I Clinical Trial for Melanoma:

Intralesional Targeted Alpha Therapy

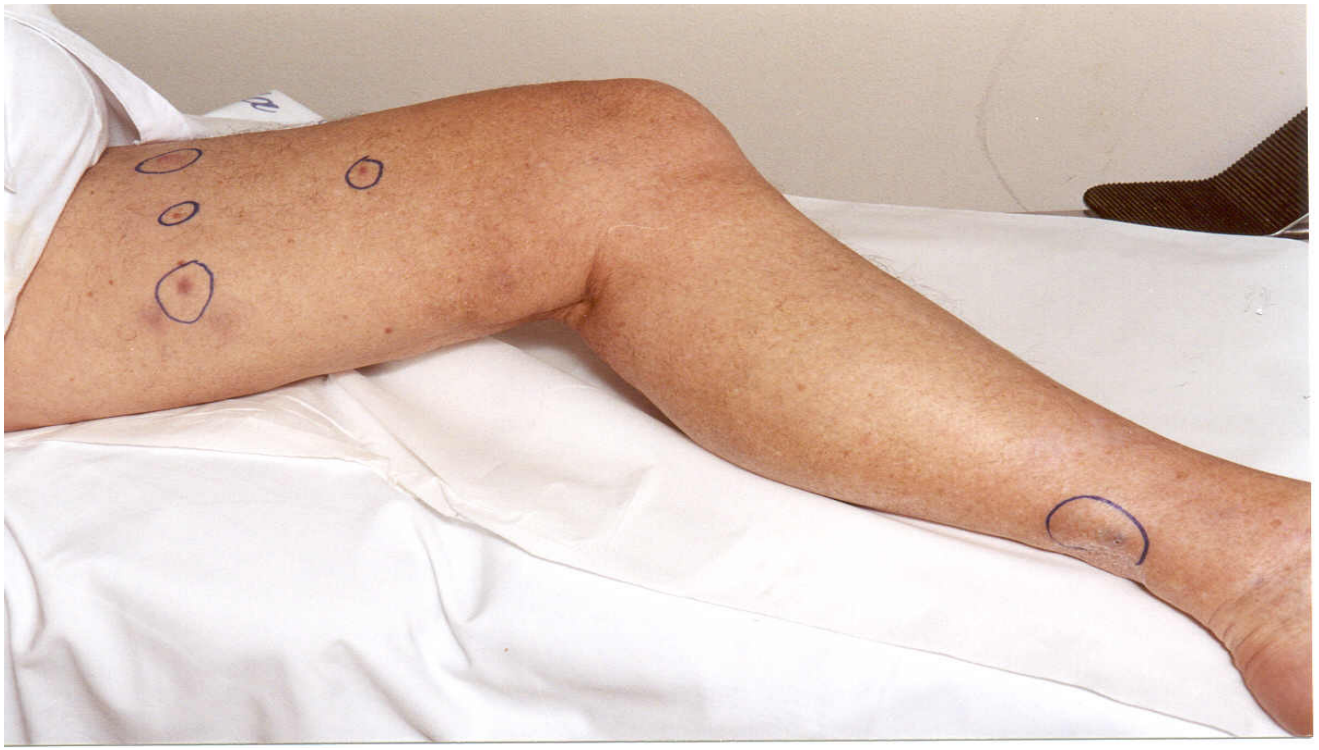
Collaboration ITU - Centre for Experimental Radiation Oncology
St George Hospital, Kogarah, Australia

- 16/16 melanomas were positive to the antibody 9.2.27mAb
- No complications observed in 16 patients treated with doses up to 1.35 mCi.
- Histology of excised tumours shows extensive cell killing

=> Intralesional target alpha therapy is safe and effective for local melanoma and may control the progression of melanoma

B. J. Allen, C. Raja, S. Rizvi, Y. Li, W. Tsui, P. Graham, J. F. Thompson, R. A. Reisfeld, J. Kearsley, A. Morgenstern, C. Apostolidis: Intralesional targeted alpha therapy for metastatic melanoma. Cancer Biol. Ther. 4(12) (2005) 1318 – 1324

=> Phase I Clinical trial of systemic application ongoing (27 patients so far)



20 of 21 melanomas on the leg of one patient completely regressed after administration of 7 mCi ^{213}Bi-9.2.27, with no local recurrences at 12 months post-treatment. This result is shown for the larger tumours, where the estimated original tumour size is shown as a blue ring.

C. Raja, P. Graham, S. Rizvi, E. Song, H. Goldsmith, R. Smart, P. Butler, J. Thompson, A. Bosserhoff, A. Morgenstern, C. Apostolidis, J. Kearsley, R. Reisfeld, B. J. Allen: Therapeutic responses in systemic targeted alpha therapy trial for melanoma. 2006, *submitted*

Treatment of Non-Hodgkin's Lymphoma

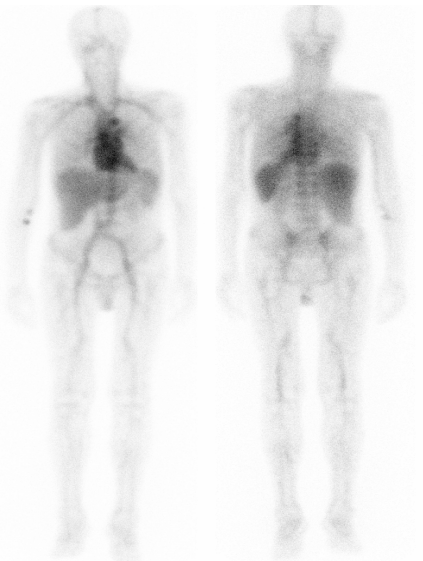
Phase I (dose escalation) study (ITU - DKFZ / UH Düsseldorf)

Previous study: German Cancer Research Center (DKFZ, Heidelberg, Germany)

- 9 patients with B-cell-malignancy
- 15-45 mCi 213Bi-antiCD20
- no toxicity observed

Study continues at University Hospital Düsseldorf, Germany

- 3 patients in 2005 (13, 52, 58 mCi)
- mild toxicity observed
- bone marrow as dose limiting organ



Bi-213 gamma emission (440 keV) allows imaging of bio-distribution

D. Schmidt, F. Neumann, C. Antke, C. Apostolidis, S. Martin, A. Morgenstern, R. Molinet, S. Heeger, R. Kronenwett, H. W. Müller and R. Haas: Phase I clinical study on alpha-therapy for Non Hodgkin Lymphoma In: Proc. 4th Alpha-immunotherapy symposium, Morgenstern, A. Ed.; Düsseldorf, Germany, June 28-29, 2004.

Equipment for Ac-225/Bi-213 generator elution and antibody labeling at University Hospital Düsseldorf

Alpha-Immunotherapy of brain tumours

Collaboration ITU - University Hospital Basel, Switzerland

- In 2000 two brain tumor patients (life expectancy approx. 6 months) treated
- application after brain tumour resection into surgically created cavity to destroy residual tumour tissue
- Bi-213 bound to oligopeptide DOTAGA-Substance P

Outcome

One patient did not respond
Second patient is still alive (>5 years),
recent MRI scan did not detect any residual tumor

Further treatments planned in 2007

Kneifel S, Cordier D, Good S, Ionescu MC, Ghaffari A, Hofer S, Kretzschmar M, Tolnay M, Apostolidis C, Waser B, Arnold M, Mueller-Brand J, Maecke HR, Reubi JC, Merlo A.: Local targeting of malignant gliomas by the diffusible peptidic vector 1,4,7,10-tetraazacyclododecane-1-glutaric acid-4,7,10-triacetic acid-substance P. Clin Cancer Res. 2006 Jun 15;12(12):3843-50

Joint Research Centre

Alpha-Immunotherapy of leukemia

ITU + Memorial Sloan Kettering
Cancer Center, New York

Phase I:

- 18 patients with relapsed and refractory AML (acute myelogenous leukaemia) or CML (chronic myelomonocytic leukaemia)
- 0.3 - 1.0 mCi/kg body weight of ^{213}Bi-HuM195 (anti-CD33)
- no significant toxicity
- 14/18 patients responded

Jurcic JG, Larson SM, Sgouros G, McDevitt MR, Finn RD, Divgi CR, Ballangrud AM, Hamacher KA, Ma D, Humm JL, Brechbiel MW, Molinet R, Scheinberg DA.: Targeted alpha particle immunotherapy for myeloid leukemia. Blood. 2002 Aug 15;100(4):1233-9.

Phase I/II (ongoing):

- elimination of residual disease after partial cytoreduction using chemotherapy
- 1.0 - 1.25 mCi/kg body weight of ^{213}Bi-HuM195
- >30 patients treated so far, approx. 50 % showed response

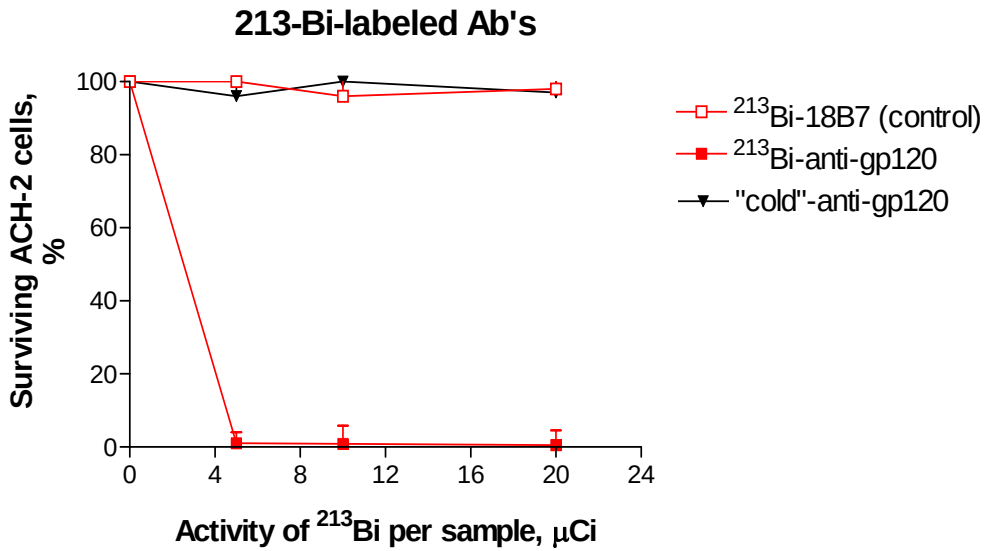
First AIT drug to be approved by FDA?

Joint Research Centre

Radioimmunotherapy of Fungal, Bacterial and Viral Infections

ITU + Albert Einstein College of Medicine, NY, USA

Targeting and elimination of HIV-infected cells *in vitro* and *in vivo*



- treatment of infected mice with ^{213}Bi-246D effectively reduced number of HIV1-infected cells by 300-fold
- no acute toxicity in treated mice

=> Study suggests Alpha-Immunotherapy as novel option for therapy of HIV infections in combination with conventional antiretroviral therapy (HAART)

E. Dadachova, M. Patel, S. Toussi, C. Apostolidis, A. Morgenstern, M. W. Brechbiel, M. K. Gorny, S. Zolla-Pazner, A. Casadevall, H. Goldstein: TARGETED KILLING OF VIRALLY-INFECTED CELLS BY RADIOLABELED ANTIBODIES TO VIRAL PROTEINS. Public Library of Science Medicine, 2006, *in press*

Joint Research Centre

Summary

- Alpha-Immunotherapy using Bi-213 is a novel and promising treatment strategy

- Alpha emitters linked to tumour selective carrier molecules have the potential for highly effective and specific killing of cancer cells

- Currently approx. 10 different types of cancer as well as infectious diseases are being studied

- Worldwide clinical trials are underway for the treatment of leukaemia, Non-Hodgkins Lymphoma, brain tumours, metastatic melanoma (total > 100 patients treated)